Molecular Targets in Metastatic Colorectal Cancer: A Review

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Abstract

In recent years, the molecular and genetic features of colorectal cancer (CRC) have been used to categorize the disease, which has made it possible to develop therapeutic approaches based on predictive biomarkers. Valuable drivers for individualized treatment plans are biomarkers including *NTRK* fusions, *RAS* and *BRAF* mutations, HER2 amplification, and microsatellite instability (MSI). Furthermore, the regular use of molecular predictive diagnostics, including liquid biopsies and the reintroduction of anti-epidermal growth factor receptor (EGFR) monoclonal antibodies, presents new opportunities for the therapeutic management of patients with CRC. With an emphasis on recent developments in EGFR blockade and novel biomarkers (MSI, HER2, and NTRK), we have provided an overview of the state of targeted therapy for patients with metastatic CRC in this review.

Keywords
- colon
- neoplasm
- Ras
- Braf
- ctDNA

Introduction

Colorectal cancer (CRC) is the second cause of death globally and the third most common type of neoplasm.[1] When molecular targeted therapy and chemotherapy are combined, the median overall survival (OS) for patients with metastatic disease is between 25 and 30 months.[2]

Surgery and chemotherapy are the backbones of treatment for localized CRC. The development of biomarkers for targeted therapies, such as immune checkpoint inhibitors (ICIs): epidermal growth factor receptor (EGFR) inhibitors, BRAF inhibitors, HER2 inhibitors, or NTRK inhibitors, have improved therapeutic strategies in metastatic setting. *RAS* and *BRAF* mutations, microsatellite instability (MSI), and mismatch repair deficiency (dMMR), *HER2* amplifications, and *NTRK* fusions are now predictive indicators for patients with metastatic disease.

In this review, we examine the latest predictive biomarkers for patients with metastatic CRC (mCRC) and the new targeted therapy that include new developments for cancers with BRAFV600E mutation, anti-HER2 therapies, NTRK inhibitors, and emerging issues for anti-EGFR agents, such as primary tumor sidedness (PTS) and longitudinal follow-up using circulating tumor deoxyribonucleic acid (ctDNA).

Materials and Methods

We have searched PubMed (www.ncbi.nlm.nih.gov/pubmed) for full-text articles published from 2017 to January 31, 2025, using the keywords "colon," "neoplasm," "RAS," "BRAF," and "ctDNA." The full-text articles found were carefully examined. In addition, all abstracts presented at international conferences between January 2020 and January 2025 were reviewed.

Anti-EGFR Therapy and RAS/RAF Wild-Type mCRC

Predictive Drivers of Anti-EGFR Agent Effectiveness

Anti-EGFR resistance in CRC patients is caused by activating mutations of *KRAS* and *NRAS*.[3] Thus, 40 to 50% of patients with CRCs have a *KRAS* mutation, while 4 to 8% have an *NRAS*

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mutation.[4] *KRAS* exons 2, 3, and 4 (codons 12, 13, 59, 61, 117, and 146) and *NRAS* exons 2, 3, and 4 (codons 12, 13, 59, 61, and 117) are recommended for *RAS* mutation testing before starting any treatment in metastatic setting.[5,6] Anti-EGFR-targeting therapies are available for patients with KRAS/NRAS wild-type (WT).

There exists additional mechanism of resistance, like the mutations of the EGFR ectodomain that may implicate ineffectiveness of anti-EGFR.[7] In addition, although the BRAFV600E mutation was not officially shown to be a cause of resistance to anti-EGFR (see below), it may also be connected to the overactivation of a protein downstream from the EGFR in the mitogen-activated protein kinase (MAPK) pathway.[8] Monoclonal antibody (mAb) resistance may be a result of constitutional activation of the PI3K/Akt/mTOR pathway by *PIK3CA* exon 20 mutation or *PTEN* deletion.[9,10] Also, resistance to anti-EGFR therapy appears to be linked to amplifications of HER2, HER3, or MET and HER2-activating mutations.[11] Finally, the predictive significance of the microRNA miR-31-3p was recently revealed. The RAS signaling pathway is largely activated by Mir-31, and elevated expression of miR-31-3p may be an indication of the tumor's EGFR independence and, hence, its resistance to anti-EGFR. Numerous post hoc analyses of randomized trials demonstrated that miR-31-3p expression is a reliable indicator of anti-EGFR effectiveness.[12–14]

Management of Anti-EGFR Therapy

In adjuvant setting, resected stage III colon cancer, anti-EGFR mAbs do not improve outcomes.[15] The NEW EPOC study raises concerns about the use of anti-EGFR mAbs in the perioperative setting for patients with resectable liver metastasis in mCRC. According to this study, cetuximab is detrimental to OS and disease-free survival when combined with chemotherapy.[16] Anti-EGFR mAbs may be useful as a converting therapy to reduce resectable metastatic disease; however, they should not be used as a perioperative treatment for patients with resectable mCRC.[17]

Cetuximab and panitumumab, two anti-EGFR mAbs largely used in clinical practice, have been linked to better response rates, OS, and progression-free survival (PFS) in first-line mCRC, in combination with regimens based on oxaliplatin or irinotecan, as well as in second or later lines alone or in combination with chemotherapy.[18–29] Recent data from the phase III study TAILOR reveal that cetuximab can be safely added to FOLFOX for RAS WT mCRC patients,[30] even though NORDIC VII and COIN studies did not demonstrate a meaningful effect of cetuximab in combination with an oxaliplatin-based regimen.[31,32] Except for chemoresistant disease, where the ASPECCT study demonstrated the noninferiority of panitumumab compared to cetuximab in patients with chemotherapy-refractory KRAS WT (exon 2) mCRC, there is no direct comparative study between cetuximab and panitumumab.[30,33]

The Role of the Sidedness

Anti-EGFR activity appears to be determined by PTS. There is mounting evidence that PTS predicts responsiveness to anti-EGFR mAbs and it is a prognostic factor in RAS WT population.[34] A retrospective study of six randomized trials (CRYSTAL, FIRE-3, CALGB 80405, PRIME, PEAK, and 20050181) revealed that right-sided colon cancer had worse outcomes (OS, PFS, and response rates) than left-sided tumors. In patients with left-side mCRC, this meta-analysis demonstrated a predictive role of PTS. Indeed, chemotherapy plus anti-EGFR mAbs had a better outcome than chemotherapy with bevacizumab in left side mCRC.[35] The predictive role of PTS was limited to the KRAS WT population, according to a recent retrospective analysis of the ARCAD database. This analysis also validated the predictive role of PTS for cetuximab efficacy, with better results for patients with left-sided mCRC.[36] Conversely, anti-EGFR therapies appear to have a worse effect on patients with RAS WT right side mCRC.

Due to their retrospective nature, these results should be interpreted carefully, but they indicate that anti-EGFR mAbs plus chemotherapy should only be used as first line for patients with left-sided tumors KRAS/NRAS WT and that patients with right-sided mCRC may benefit more from chemotherapy plus an antiangiogenic agent.[37]

Rechallenge and Liquid Biopsy

Tumor clones with an intrinsic mutation of resistance are selected during treatment with anti-EGFR, causing acquired resistance to this drug. The tumor can recover sensitivity when the anti-EGFR mAb is discontinued, since this removes the positive pression selection on the sensitive clones. Tumor resistance can be overcome by a variety of methods, including reintroduction, dose intensification, sequential therapy, and rechallenge; in the case of anti-EGFR mAbs, rechallenge, this strategy appears to be the most promising.[38] For a tumor that first showed sensitivity to anti-EGFR therapy, retreatment following a progression could be referred to as a challenge of anti-EGFR therapy.[39]

For rechallenge strategy, longitudinal follow-up of mutant clones is interesting. According to studies using longitudinal ctDNA monitoring, RAS mutant clones developed in blood during anti-EGFR therapy have a half-life of 4 to 5 months before declining rapidly after end treatment.[40] The first prospective trial that demonstrated that a rechallenge strategy using cetuximab and irinotecan might be effective in RAS/BRAF WT mCRC patients with acquired resistance to cetuximab was the CRICKET phase II study. Blood samples from patients who reported partial response did not show any RAS mutation.[41,42] The utility of liquid biopsy in the context of anti-EGFR rechallenge was assessed in several clinical trials (i.e., CHRONOS, RASINTRO) that demonstrated the same results.[41]

Braf Mutation in mCRC

About 8 to 10% of mCRC exhibit *BRAFV600E* mutation, which causes a *RAS*- independent constitutional activation of the MAPK pathway promoting cell survival and proliferation and being linked to a poor prognosis.[43] While 22% of all *BRAF* mutations in CRC occur outside of the V600E hotspot, these mutations do not have the same biochemical, clinical, and therapeutic effects as the V600E mutation.[44] Although some

may be responsive to EGFR, these BRAF non-V600E mutant tumors are more likely to be left-sided, have a lower grade of differentiation, and have a better prognosis. They are also resistant to BRAF inhibitors.[45,46] These genetic changes appear to not provide resistance to anti-EGFR therapy and are linked to malignancies on the right side.[47,48]

Patients with BRAFV600E CRC are more likely to be older, female, and have right-sided tumors with a mucinous component. Furthermore, these patients are also most prone to have distant lymph node and peritoneal metastases, but fewer pulmonary metastases.[49] Significantly, the MSI phenotype, which is indicative of the effectiveness of ICIs regardless of the BRAF mutational status, is present in around 22% of BRAFV600E mCRC.[50]

Compared to BRAF WT, BRAFV600E-mutated mCRC are less likely to get second- line therapies. Intensification therapies appear to work well for these patients.[51–53] Compared to FOLFIRI (folinic acid, fluorouracil, and irinotecan) plus bevacizumab, first-line FOLFOXIRI (folinic acid, fluorouracil, oxaliplatin, and irinotecan) plus bevacizumab was linked to a nonsignificant improvement in OS for BRAFV600E mutants in the TRIBE study.[54] For patients with BRAFV600E mCRC chemotherapy-naive, FOLFOXIRI-bevacizumab is regarded as a viable treatment choice, notwithstanding the limited population sample included in this subgroup analysis. Crucially, a subgroup analysis on 33 patients BRAF Mut V600E in the TRIBE2 phase III trial, which compared mFOLFOX6 plus bevacizumab followed at progression from FOLFIRI plus bevacizumab like TML strategy, with FOLFOXIRI plus bevacizumab stop and go did not reveal any survival benefit for BRAFV600E patients.[55] The Fire 4.5 study (AIO-KRK-0116) phase II trial evaluated the triplet chemotherapy regimen with either cetuximab or bevacizumab (NCT04034459; see ►**Table 1**). The primary endpoint objective response rate (ORR) was on experimental arm of 51% and in the control arm of 61%.

Braf V600E Mutations and Antiangiogenic Drugs

To date, there are no studies that have demonstrated predictive markers for antiangiogenic drugs, and their efficacy in BRAFV600E mCRC patients has not been demonstrated. Adding bevacizumab to first-line IFL (bolus irinotecan, fluorouracil, and folinic acid) or capecitabine did not increase survival, according to the AVF2107 and AGITG MAX36 studies.[56,57] Although the limited size of the patients did not allow the evaluation of statistical significance, the VELOUR trial (FOLFIRI ± aflibercept) and the RAISE study (FOLFIRI plus ramucirumab) demonstrated that patients with BRAF V600E mutations tended to benefit from the antiangiogenic drugs in second line.[58,59] All things considered, this retrospective analyses imply that antiangiogenics in first line may be helpful for patients with BRAFV600E mCRC.[60]

Anti-EGFR and BRAFV600E Mutations

It is unclear if anti-EGFR treatments, either alone or with chemotherapy, are effective for BRAFV600E patients. There were two meta-analyses conducted. According to a meta-analysis by Pietrantonio et al, patients with BRAFV600E do not respond well to anti-EGFR drugs.[61] However, no discernible difference in the impact of anti-EGFR drugs between the BRAFV600E and BRAF WT populations was seen in another meta-analysis conducted by Rowland et al.[62] Furthermore, the FIRE-3 study (first-line FOLFIRI plus cetuximab vs. FOLFIRI plus bevacizumab in KRAS WT mCRC patients) revealed a greater response rate in the cetuximab arm, according to a retrospective analysis of the BRAFV600E subgroup.[63] Also, the subset of BRAFV600E patients showed a significant increase in objective response (71% vs. 22%, $n = 14$) in a recent study (VOLFI AIO KRK0109) evaluating the effectiveness of first-line FOLFOXIRI with or without panitumumab.[18] However, despite the conflicting data, the European Society for Medical Oncology and National Comprehensive Cancer Network guidelines do not recommend the first-line use of anti-EGFR in patients with Braf V600E mutated mCRC.

Inhibitors of BRAF

Unlike melanoma, BRAF inhibitors in mCRC alone were linked to unsatisfactory outcomes. One theory is that BRAF inhibition may encourage MAPK constitutive signaling by triggering feedback EGFR activation. One factor contributing to these cancers' innate resistance to BRAF inhibitor monotherapy is the EGFR-mediated reactivation of downstream signaling cascades.[64,65] Several combinations of BRAF inhibitors, anti-EGFR, PI3K inhibitors, or MEK inhibitors were explored with thisproblem in mind, and the findings were

Table 1 Ongoing clinical trials for patients with *BRAF*V600E metastatic colorectal cancer

Therapy	Phase	Condition	Primary endpoint	NCT identifier
Encorafenib[1] + cetuximab[2] + nivolumab[4]	1/2	2nd or 3rd line	ORR, DLT	NCT04017650
Encorafenib[1] + binimetinib[3] + nivolumab[4]	1/2	> 1st line	ORR, DLT	NCT04044430
Dabrafenib[1] + trametinib[3] + PDR 001[4]	2	Any line	ORR, DLT	NCT03668431
FOLFOXIRI + cetuximab[2] or bevacizumab[5]	2	1st line	ORR	NCT04034459
FOLFIRI + cetuximab[2] + vemurafenib[1]	2	–	ORR	NCT03727763
Irinotecan + AZD 1775[6]	1	> 1st line	DLT	NCT02906059
Panitumumab[2] + trametinib[3]	2	> 2nd line	ORR	NCT03087071

Abbreviations: DLT, dose-limiting toxicities; EGFR, epidermal growth factor receptor; NCT, National Clinical Trial; ORR: objective response rate; VEGF, vascular endothelial growth factor.
Note: [1]RAF inhibitor; [2]EGFR inhibitor; [3]MEK inhibitor; [4]anti-PD(L)-1; [5]anti-VEGF; [6]Wee-1 inhibitor.

intriguing.[66–70] These studies provided support for the design of the phase III BEACON, which compared chemotherapy (investigator choice regimen of cetuximab plus irinotecan or FOLFIRI) with encorafenib and cetuximab ± binimetinib. Randomization was performed on 665 BRAFV600E mCRC patients whose disease had progressed after one or two prior lines of chemotherapy. In the triplet and doublet experimental arms, the median OS was 9.3 months, while in the control arm, it was 5.9 months (hazard ratio [HR] = 0.60, 95% confidence interval [CI] 0.47–0.75 and HR = 0.61, 95% CI 0.48–0.77, respectively).[64,71] A statistical improvement was observed in the ORR, which was 2% in the control group and 20 and 26% in the doublet and triplet arms, respectively. The experimental groups experienced cutaneous and gastrointestinal side effects, but the toxicity was tolerable, with grade 3 or higher toxicities being similar across the three arms. Both the doublet and triplet groups had a lower chance of quality-of-life decline by over 40%, according to a supplemental quality-of-life analysis.

Recently was presented at ASCO GI 2025 the abstract of Breakwater study, a phase III that compares first-line Braf Mut V600E mCRC, encorafenib plus cetuximab and FOLFOX versus SOC. The primary endpoint, ORR, was met with a ORR of 60.9% for the experimental arm and 40.0% ($p = 0.0008$) for the control arm.[72]

Targeted Therapies in Patients with Ras Mutations

KRAS/NRAS mutations are present in more than 50% of patients with mCRC. As shown above, they are inherently resistant to anti-EGFR mAbs. Although there are no predictive biomarkers for the effectiveness of antiangiogenics (bevacizumab, aflibercept, and ramucirumab), these drugs appear to be beneficial in this population.[59,73,74]

One of the mutations for which a drug target is being studied is G12C (glycine 12 to aspartic acid). For this population, a novel class of KRAS inhibitors may prove revolutionary.[75] In the phase III Codebreak 300 study, AMG 510 (sotorasib) was administered in later lines to patients with G12C mutation mCRC. The study included three arms. The first enrolled patients with sotorasib 960 mg with panitumumab, the second arm sotorasib 240 mg with panitumumab. In the third arm, patients were started on treatment with TAS 102 or regorafenib at the investigator's choice. The primary endpoint was PFS.

After a median follow-up of 7.8 months, PFS was 5.6, 3.9, and 2 months, respectively. The statistical comparison between the first arm and the third arm was statistically significant in favor of the experimental arm (95% CI, 0.30–0.78; $p = 0.005$).[76]

Immune Checkpoint Inhibitors and Microsatellite Instability

Microsatellite Instability, Mismatch Repair Deficiency, and Colorectal Cancers

From 10 to 15% of CRCs originate from the MSI pathway, the majority grow through the chromosomal instability pathway (aneuploidy and loss of genetic material). A germline mutation in the MMR genes (*MLH1*, *PMS2*, *MSH2*, *MSH6*) that predispose to Lynch syndrome or an epigenetic inactivation of MLH1 (i.e., sporadic malignancies) results in a deficiency of the DNA dMMR pathway, so-called MSI. The BRAFV600E mutation is commonly linked to these isolated occurrences.[77] About 10 to 15% of localized CRC and 4 to 5% of mCRC at the fourth stage, have MSI/dMMR.[43,78] The right colon is the primary site of origin for MSI/dMMR CRCs, which exhibit distinct characteristics such as low differentiation, a high number of tumor-infiltrating lymphocytes, and characteristic metastatic patterns, including frequent distant lymph node metastases and peritoneal involvement.[49] MSI/dMMR is linked to a good prognosis in localized CRC.[79,80] In metastatic disease, data are more controversial. However, the existing trials indicates that, in comparison to microsatellite stable/MMR-proficient (MSS/pMMR) cancers, MSI/dMMR mCRC are less susceptible to traditional treatment.[81–83]

High tumor mutational burden (hypermutated phenotype) and highly immunogenic neoantigens resulting from frameshift mutations that cause high infiltration through activated cytotoxic T CD8+ cells are characteristics of MSI/dMMR CRCs.[84–86] Nevertheless, immunological checkpoints are upregulated in MSI/dMMR cancers, shielding MSI cancer cells from their tough immune environment.[87,88]

Immune System as a Target of Therapy

For patients with mCRC, MSI/dMMR has become a considerable prognostic biomarker for the effectiveness of ICIs. MSI/dMMR cancers were linked to significant sensitivity to immunotherapy (i.e., hot tumors), whereas MSS/pMMR CRCs are mostly resistant to ICIs (i.e., cold tumors). Several phase II trials have shown that ICIs are effective for patients with chemoresistant MSI/dMMR mCRC, with ORRs ranging from 33 to 58% and 12-month PFS rates between 31 and 71%.[50,89–94] Anti-PD1 and anti-CTLA4 mAb combinations may be more effective than anti-PD1 or anti-PDL1 alone, according to the findings of the nonrandomized CheckMate-142 trial. Indeed, in a third cohort of the CheckMate-142 study, 45 patients received nivolumab + ipilimumab in first-line chemotherapy-naive MSI/dMMR mCRC, demonstrating the effectiveness of ICIs as front-line treatment. The 1-year PFS estimate was 77%, and the ORR was 77%.[95] Another trial, the phase III KEYNOTE 177, demonstrated in fist line that pembrolizumab monotherapy had better PFS in MSI/dMMR mCRC patients compared to standard-of-care (investigator's choice of FOLFOX or FOLFIRI, with or without bevacizumab or cetuximab). The primary endpoint, median PFS, were 16.5 and 8.2 months (HR = 0.60, 95% CI 0.45–0.80). With pembrolizumab, the 12- and 24-month PFS rates were 55 and 48%, respectively, while with chemotherapy, they were 37 and 19%. For patients with newly diagnosed MSI/dMMR mCRC, pembrolizumab has become the standard of therapy.[96]

For patients with localized MSI/dMMR colon cancer, ICIs are presently being assessed. Their development in this context was made possible by the NICHE phase II trial, which may also improve treatment approaches for MSI/dMMR CRC in its early stages.[97] All 21 dMMR CRC patients experienced a

pathological response in this trial evaluating nivolumab with ipilimumab as a neoadjuvant treatment; 12 full pathological responses were among the 95% of major responses. These remarkable outcomes demonstrate that neoadjuvant immunotherapy is a viable approach that merits more investigation. In the ATOMIC trial (NCT02912559; FOLFOX ± atezolizumab) and the POLEM trial (NCT03827044; 24 weeks of single agent fluoropyrimidine chemotherapy or 12 weeks of oxaliplatin-based chemotherapy ± avelumab), ICIs are also assessed in conjunction with adjuvant chemotherapy for patients with stage III MSI/dMMR colon cancer.[28]

Predictive Biomarkers in Immunotherapy

MSI/pMMR patients respond to ICIs for a short period and then develop resistance to them. No other biomarkers are known to predict response to immunotherapy in this cohort of patients. Interestingly, a considerable number of cases with primary resistance to ICIs are caused by misinterpretation of MSI/dMMR status.[98,99]

The patients with tumors MSI/dMMR BRAF WT appear to be highly sensitive to ICI as the patients with MSI/dMMR, BRAFV600E mutated.[50] The resistance to ICI was not linked to major histocompatibility complex class I expression, beta-2-microglobulin mutations, or PD-1 expression.[100] ICI resistance in MSI/dMMR mCRC may be caused by loss-of-function mutations in Janus kinases JAK1/2.[101] Remarkably, in two small cohort trials (less than 33 patients), the tumor mutational burden was found to predict the effectiveness of ICI.[102,103] Interesting data, but not yet translatable to clinical practice, are available on the immune infiltrate. The degree of T cell infiltration was associated with improved response, PFS, and OS in a recent study by Loupakis et al.[99] Larger prospective studies should corroborate all of these findings.

HER2 and Anti-HER2

HER2 gene amplification is present between 1 and 8% of patients with CRC.[104–107] *KRAS* WT status and HER2 overexpression are linked and are more present in left mCRC, with a frequency of 4.3 to 5.4%.[108,109] To date, we know the role of HER2 as a negative prognostic factor for resistance to anti-EGFR.[110,111]

The Heracles diagnostic criteria established a standard procedure for HER2 testing in CRC, which included before immunohistochemistry (IHC) analysis and, if necessary, fluorescence in situ hybridization (FISH). An IHC 3+ score or an IHC 2+ score linked to FISH positivity is used to define positivity.[112]

The effectiveness of anti-HER2 drugs for patients with HER2-positive mCRC is verified. Phase II studies evaluated trastuzumab with lapatinib, trastuzumab plus pertuzumab, and trastuzumab plus tucatinib (Heracles-A, MyPathway, and Mountaneer, respectively). The median PFS was 4.7, 2.9, and 6.2 months, respectively, and response rates were 30, 32, and 55%.[113,114] The Mountaneer and Heracles-A studies did not include patients with HER2-positive and KRAS-mutated mCRC; nevertheless, it is noteworthy that one patient with HER2-positive and KRAS-mutated mCRC had an objective response in the MyPathway study.[113,114] The Heracles-B study, which involved the combination of pertuzumab and trastuzumab emtansine, did not achieve its primary endpoint (ORR) but had a median PFS of 4.7 months.[115] According to a recent study from the DESTINY-CRC01 phase II trial, trastuzumab–deruxtecan may change the future. This antibody drug conjugated, which consists of a topoisomerase I inhibitor and an anti-HER2 antibody, was used to treat 50 patients with chemoresistant HER2-positive mCRC. A confirmed ORR of 45% was obtained. With an ORR of 43.8%, this treatment was beneficial even for individuals who had previously used anti-HER2 drugs. Two patients succumbed to interstitial lung disease due to the drugs.

Although randomized trial data are insufficient for a thorough assessment of the additional value of anti-HER2, these drugs are generally very appealing treatments for the HER2-positive population. In patients with HER2-positive RAS/RAF WT mCRC, the only randomized study currently in progress is a phase II trial that compares trastuzumab and pertuzumab to cetuximab and irinotecan (SWOG S 1613 NCT03365882).

TRK Inhibitors and NTRK Gene Fusions

Recently, NTRK gene fusions have become a very appealing therapeutic target for cancer patients. Regardless of the histology type, TRK inhibitors (entrectinib, larotrectinib) showed remarkable therapeutic activity in various types of cancers. In single-arm trials, entrectinib had an ORR of 57% with a time of response greater than 6 months in 68% of patients, and larotrectinib demonstrated an ORR of 75% with a time of response greater than 6 months in 73% of cases.[116,117] Due to these findings, the Food and Drug Administration has arranged a fast-track approval for the use of the NTRK gene fusion to treat refractory solid tumors, regardless of the kind of tumor.

Depending on the likelihood of NTRK fusion, screening methods for this mutation rely on next-generation sequencing, reverse transcription polymerase chain reaction, and immunohistochemical FISH.[118,119] With an incidence of 0.23 to 0.97%, NTRK fusions are uncommon in CRCs.[120–123] Females, right-sided initial tumor site, RAS/RAF WT status, and MSI phenotype are characteristics of individuals with CRC that have NTRK fusion.[121] Interestingly, NTRK fusions were consistently linked to the MSI phenotype. More specifically, hypermethylation of the MLH1 gene promoter appeared to be associated with these genetic changes in BRAF WT tumors.[124,125] In this molecularly chosen sample, the estimated incidence of NTRK fusions was 42%.[48] The effectiveness of ICIs and NTRK inhibitors in this particular biological entity is not yet known.

Conclusion

Over the past 10 years, notable progress has been achieved in tailoring treatment plans for patients with mCRC. An expanded panel of biomarkers can be used to specifically

Table 2 Molecular subtypes of colorectal cancer and targeted treatment options

Molecular subtypes	Targeted therapies
MSI, whatever the *RAS*/*RAF* mutational status	Immune checkpoint inhibitor(s)
RAS/*RAF* wild-type	Anti-EGRF mAbs
*BRAF*V600E mutated	Encorafenib + cetuximab ± binimetinib
RAS mutated	No current targeted therapy, ongoing trials with new-generation KRAS inhibitors
HER2 amplified/mutated	Anti-HER2 mAbs/inhibitors (trastuzumab, pertuzumab, lapatinib), anti-HER2 antibody-drug conjugate (trastuzumab deruxtecan)
NTRK fusion-positive	TRK inhibitor (larotrectinib, entrectinib)

Abbreviations: mAb, monoclonal antibody; EGFR, epidermal growth factor receptor; MSI, microsatellite instability; TRK, tropomycin receptor kinase.

identify responders to anti-EGFR therapy, and ctDNA longitudinal follow-up can be used to optimize therapeutic approaches. Previously untreated patients with BRAFV600E mCRC now have access to efficient treatment alternatives. Beyond extremely attractive but extremely uncommon targets like NTRK fusions and HER2 amplification, ICIs—a breakthrough for patients with MSI/dMMR tumors—have brought about the most notable change in targeted therapy for patients with CRC. Because of the significant improvement in patient outcomes, researchers and clinicians were forced to consider CRC as at least two different diseases: the MSI/dMMR tumors and the rest (►**Table 2**). Crucially, methodological problems with the pseudoprogression phenomena and long-term survivals are linked to the creation of ICIs. This finding emphasizes the need to create novel study designs and to account for these problems in statistical analyses that are planned in the future.

Patient Consent
Patient consent is not required.

Conflict of Interest
None declared.

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